STATE OF MARYLAND

DHMH

Maryland Department of Health and Mental Hygiene

Larry Hogan, Governor - Boyd Rutherford, Lt. Governor - Van Mitchell, Secretary

Laboratories Administration
Robert A. Myers, Ph.D., Director

DATE: February 2, 2016

TO: Medical Laboratory Directors, Local Health Officers, Health Care Providers

FROM: Robert A. Myers, Ph.D. RAM
Director, Laboratories Administration

Maria Paz Carlos, Ph.D MPC
Chief, Division of Virology and Immunology, Laboratories Administration

RE: Interim Guidance and Instructions for Submission of Specimens for Suspected Zika Virus Infection Testing at CDC

In response to the emergence of Zika virus in the Americas and elsewhere and recent reports of travel-associated cases in the United States, the Maryland DHMH Public Health Laboratory will be facilitating and coordinating the submission of specimens for Zika virus testing to the Centers for Disease Control and Prevention (CDC) Laboratories.

At this time, assays to diagnose Zika virus infections are being performed at CDC Laboratories as restricted tests. We expect to develop capacity for Zika PCR testing at the MD DHMH Laboratories Administration soon. However, in the meantime, all specimens approved for testing will be tested at CDC. **Before specimens for Zika virus testing can be submitted to the MD DHMH Laboratories Administration for shipment to the CDC Laboratories, a consultation by a DHMH epidemiologist must be conducted**. This consultation will determine if the suspect case meets the clinical and travel criteria that would qualify the patient for testing. Contact your local health department (LHD) or the DHMH Infectious Disease Epidemiology and Outbreak Response Bureau at (410) 767-6700 (or after hours, at (410) 795-7365) to arrange for a consultation.

Interim guidance for assessing the suitability of persons returning from areas of ongoing transmission, including pregnant women, and infants of Zika virus infected mothers will be helpful before contacting the DHMH epidemiologist and can be found at:

- http://www.cdc.gov/zika/hc-providers/clinicalevaluation.html
- http://www.cdc.gov/mmwr/volumes/65/wr/pdfs/mm6502e1er.pdf [pregnant women]
- http://www.cdc.gov/mmwr/volumes/65/wr/mm6503e3.htm [infants]

The laboratory tests used to identify Zika virus infections primarily rely on a reverse transcriptase polymerase chain reaction (RT-PCR) to detect viral RNA in acute specimens, along with an immunoglobulin (Ig) M enzyme-linked immunosorbent assay (ELISA) and plaque reduction neutralization testing (PRNT) to identify virus specific antibodies. Please keep in mind Zika, dengue and chikungunya are mosquito-borne viruses that are being transmitted in the same geographic areas and can be difficult to distinguish clinically. Therefore, when you request testing for Zika virus, also request testing for dengue and chikungunya viruses, which will be performed.

Test Request Form:
When submitting specimens that have been approved for Zika virus testing to the DHMH Laboratory, complete the DHMH Laboratories Serological Testing Request Form No. 4677. Specimens submitted without prior approval from the LHD or the DHMH Infectious Disease Epidemiology and Outbreak Response Bureau will not be accepted for testing.

For detailed instructions for collecting and submitting acceptable specimens, please refer to the attached instructions on the DHMH Serological Testing Request Form No. 4677 Sample Form (Travel-Associated Zika Viral Infections Instructions for Specimen Submission February 2, 2016) or go to the MD DHMH Laboratories website (http://dhmh.maryland.gov/laboratories). **Please ensure that all required core demographic and contact information are completed**. In addition, please provide the following information to facilitate testing. Failure to include these additional clinical and epidemiological information might result in delays processing of specimens for testing.

a. **Name of Health Department Person Approving Testing**: Please record on the requisition the name of the DHMH or local health department person approving the testing.

b. **Clinical Illness/Compatible clinical presentation:** e.g., rash, acute onset fever, conjunctivitis, arthralgia

c. **Pertinent travel history**: Recent travel to a region where local transmission of Zika virus has been documented (an updated list is available at http://www.cdc.gov/zika/geo/index.html)

d. **History of any previous flavivirus infection:** e.g., West Nile virus (WNV), dengue virus

e. **Acute illness on-set date**: contemporaneous with the travel exposures in areas of ongoing transmission (illness on-set date ≤14 days after exposure

f. **Immunization history**: Yellow fever (YF), Japanese encephalitis (JE), or Tick-borne encephalitis (TE) vaccines

Specimen Types:
Whole blood (red-top tube/serum separator tube) or sera are acceptable specimens for both molecular and antibody testing. Molecular testing by RT-PCR will be performed on acceptable specimens collected within 7 days of the onset of illness. Antibody testing (ELISA IgM) will be performed on specimens collected ≥4 days after the on-set of illness. An additional convalescent specimen for IgM testing could be required to rule-out infections if the acute phase specimen is negative by **both** PCR and IgM testing. Extensive cross-reactivity in flavivirus serological assays has been documented. Therefore, additional PRNT testing might be performed using paired acute and convalescent sera to possibly identify the most recently infecting flavivirus.

Collect 6-10 mls of blood or 3-5 mls of serum, and transport to the lab on cold packs with completed DHMH Laboratories Serological Testing Request Form No. 4677 (see attached). (Keep specimens refrigerated until transported).

If more than 72 hours will pass to transport the specimen to MD DHMH Laboratories, serum should be separated from whole blood and frozen. The serum should then be shipped frozen.

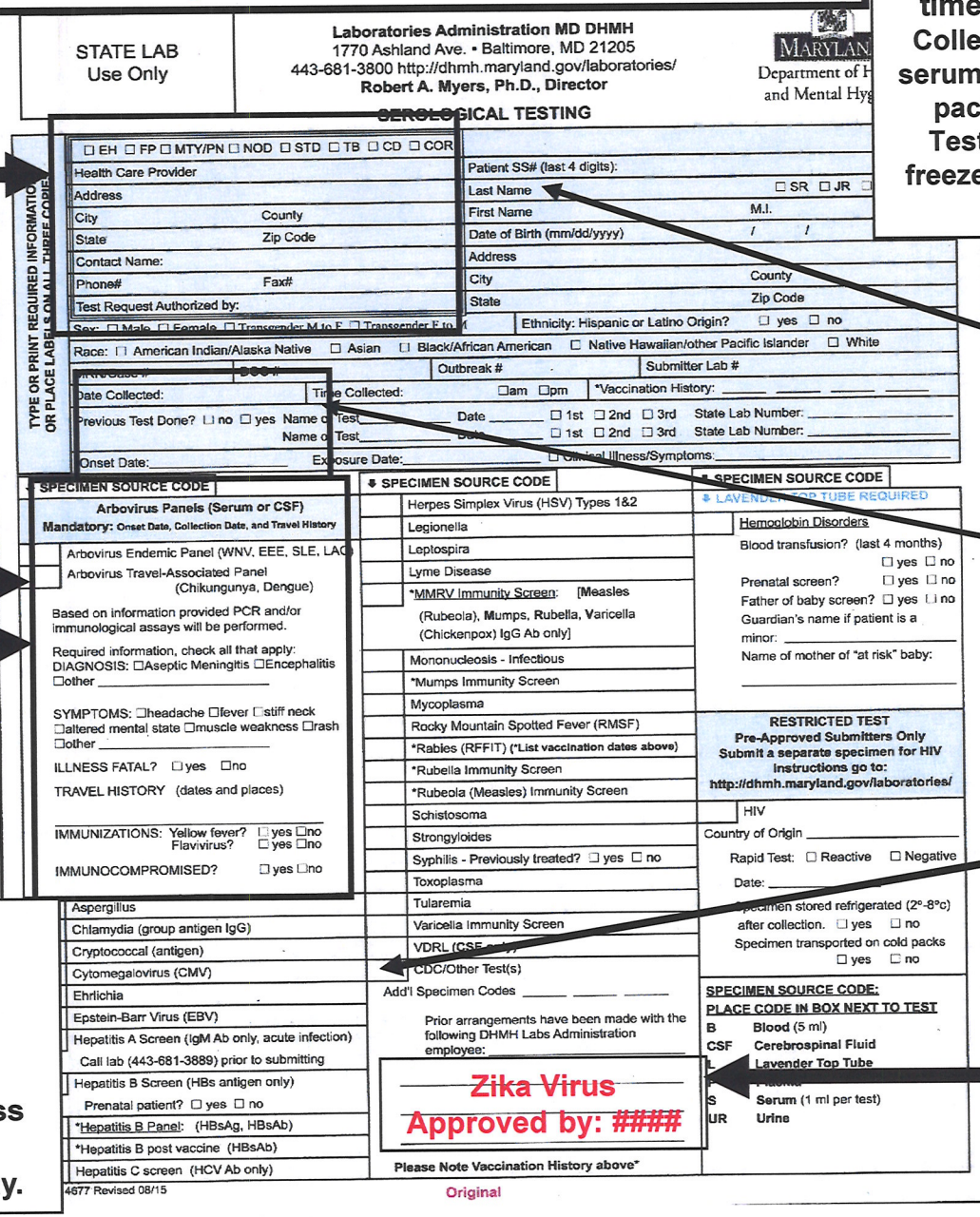
Detailed instructions on how to submit other specimen types (including amniotic fluid, cord blood, cerebrospinal fluid (CSF) and tissues) for Zika, dengue, chikungunya and other arboviral tests, contact the MD DHMH Laboratories at (443) 681-3923 or (443) 681-3937 during normal business hours from 8:00 a.m. - 4:30 p.m., Monday through Friday.

Clinical laboratories currently performing chikungunya, dengue, or in the future plan to conduct Zika virus testing, are reminded to report cases of infections and submit clinical materials (i.e. serum, CSF, etc.) from these cases to the MD DHMH Laboratories as required by statute (Annotated Code of Maryland Health-General Article, §§18-201, 18-202, and 18-205, and Code of Maryland Regulations 10.06.01.03C: #9 Arboviral Infections).

Encl:
Travel-Associated Zika Viral Infectious Instructions for Specimen Submission (February 2, 2016)
DHMH Laboratories Serological Testing Request Form No. 4677

cc: Dr. Howard Haft
Dr. David Blythe
Dr. Lucy Wilson
Dr. Richard Brooks
Dr. Katherine Feldman

Travel-Associated Zika Viral Infections
Instructions for Specimen Submission 2/2/16



STATE LAB Use Only

Laboratories Administration MD DHMH
1770 Ashland Ave. • Baltimore, MD 21205
443-681-3800 http://dhmh.maryland.gov/laboratories/
Robert A. Myers, Ph.D., Director

MARYLAND Department of Health and Mental Hygiene

SEROLOGICAL TESTING

TYPE OR PRINT REQUIRED INFORMATION OR PLACE LABELS ON ALL THREE COPIES

☐ EH ☐ FP ☐ MTY/PN ☐ NOD ☐ STD ☐ TB ☐ CD ☐ COR

Health Care Provider
Address
City County
State Zip Code
Contact Name:
Phone# Fax#
Test Request Authorized by:
Sex: ☐ Male ☐ Female ☐ Transgender M to F ☐ Transgender F to M

Patient SS# (last 4 digits):
Last Name ☐ SR ☐ JR ☐
First Name M.I.
Date of Birth (mm/dd/yyyy) / /
Address
City County
State Zip Code
Ethnicity: Hispanic or Latino Origin? ☐ yes ☐ no
Race: ☐ American Indian/Alaska Native ☐ Asian ☐ Black/African American ☐ Native Hawaiian/other Pacific Islander ☐ White

Outbreak # Submitter Lab #
Date Collected: Time Collected: ☐am ☐pm *Vaccination History: ____ ____ ____ ____
Previous Test Done? ☐ no ☐ yes Name of Test Date ☐ 1st ☐ 2nd ☐ 3rd State Lab Number:
Name of Test Date ☐ 1st ☐ 2nd ☐ 3rd State Lab Number:
Onset Date: Exposure Date: Clinical Illness/Symptoms:

SPECIMEN SOURCE CODE
Arbovirus Panels (Serum or CSF)
Mandatory: Onset Date, Collection Date, and Travel History
Arbovirus Endemic Panel (WNV, EEE, SLE, LAC)
Arbovirus Travel-Associated Panel (Chikungunya, Dengue)
Based on information provided PCR and/or immunological assays will be performed.
Required information, check all that apply:
DIAGNOSIS: ☐Aseptic Meningitis ☐Encephalitis ☐other
SYMPTOMS: ☐headache ☐fever ☐stiff neck ☐altered mental state ☐muscle weakness ☐rash ☐other
ILLNESS FATAL? ☐yes ☐no
TRAVEL HISTORY (dates and places)
IMMUNIZATIONS: Yellow fever? ☐yes ☐no Flavivirus? ☐yes ☐no
IMMUNOCOMPROMISED? ☐yes ☐no

SPECIMEN SOURCE CODE
Herpes Simplex Virus (HSV) Types 1&2
Legionella
Leptospira
Lyme Disease
*MMRV Immunity Screen: [Measles (Rubeola), Mumps, Rubella, Varicella (Chickenpox) IgG Ab only]
Mononucleosis - Infectious
*Mumps Immunity Screen
Mycoplasma
Rocky Mountain Spotted Fever (RMSF)
*Rabies (RFFIT) (*List vaccination dates above)
*Rubella Immunity Screen
*Rubeola (Measles) Immunity Screen
Schistosoma
Strongyloides
Syphilis - Previously treated? ☐ yes ☐ no
Toxoplasma
Tularemia
Varicella Immunity Screen
VDRL (CSF only)
CDC/Other Test(s)
Add'l Specimen Codes

SPECIMEN SOURCE CODE
LAVENDER TOP TUBE REQUIRED
Hemoglobin Disorders
Blood transfusion? (last 4 months) ☐ yes ☐ no
Prenatal screen? ☐ yes ☐ no
Father of baby screen? ☐ yes ☐ no
Guardian's name if patient is a minor:
Name of mother of "at risk" baby:

RESTRICTED TEST
Pre-Approved Submitters Only
Submit a separate specimen for HIV
Instructions go to:
http://dhmh.maryland.gov/laboratories/
HIV
Country of Origin
Rapid Test: ☐ Reactive ☐ Negative
Date:
Specimen stored refrigerated (2°-8°c) after collection. ☐ yes ☐ no
Specimen transported on cold packs ☐ yes ☐ no

SPECIMEN SOURCE CODE:
PLACE CODE IN BOX NEXT TO TEST
B Blood (5 ml)
CSF Cerebrospinal Fluid
L Lavender Top Tube
P Plasma
S Serum (1 ml per test)
UR Urine

Aspergillus
Chlamydia (group antigen IgG)
Cryptococcal (antigen)
Cytomegalovirus (CMV)
Ehrlichia
Epstein-Barr Virus (EBV)
Hepatitis A Screen (IgM Ab only, acute infection)
Call lab (443-681-3889) prior to submitting
Hepatitis B Screen (HBs antigen only)
Prenatal patient? ☐ yes ☐ no
*Hepatitis B Panel: (HBsAg, HBsAb)
*Hepatitis B post vaccine (HBsAb)
Hepatitis C screen (HCV Ab only)

Prior arrangements have been made with the following DHMH Labs Administration employee:

~~Zika Virus~~
~~Approved by: ####~~

Please Note Vaccination History above"

4677 Revised 08/15

Original

Must complete submitter information and include the name of the authorized person requesting the test.

Request Arbovirus Travel-Associated Panel, indicate "S"

Complete patient's travel history (locations and dates), symptoms, vaccination history, and immune status.

For questions on Zika Virus testing, please contact the lab at: 443-681-3923 or 443-681-3937 during normal business hours from 8:00AM – 4:30PM, Monday-Friday.

Whole blood (red-top/serum separator tube) or sera are acceptable specimens for both real-time PCR and IgM ELISA testing. Collect 6-10 mls of blood or 3-5 ml of serum, and transport to the lab on cold packs with completed Serological Testing form. Separate serum and freeze if held >72 hours, and transport to the lab frozen.

Patient's first and last names must be on the specimen container and match exactly to the lab slip.

Collection Date and onset date of symptoms fields **must** be completed.

Request CDC/Other Test(s), indicate "S"

You MUST write both "**Zika Virus**" to request testing and the name of the health department person who approved testing

STATE LAB
Use Only

Laboratories Administration MD DHMH
1770 Ashland Ave. • Baltimore, MD 21205
443-681-3800 http://dhmh.maryland.gov/laboratories/
Robert A. Myers, Ph.D., Director

MARYLAND
Department of Health and Mental Hygiene

SEROLOGICAL TESTING

TYPE OR PRINT REQUIRED INFORMATION OR PLACE LABELS ON ALL THREE COPIES

☐ EH ☐ FP ☐ MTY/PN ☐ NOD ☐ STD ☐ TB ☐ CD ☐ COR

Health Care Provider	Patient SS# (last 4 digits):
Address	Last Name ☐ SR ☐ JR ☐ Other_____
City County	First Name M.I.
State Zip Code	Date of Birth (mm/dd/yyyy) / /
Contact Name:	Address
Phone# Fax#	City County
Test Request Authorized by:	State Zip Code

Sex: ☐ Male ☐ Female ☐ Transgender M to F ☐ Transgender F to M

Ethnicity: Hispanic or Latino Origin? ☐ yes ☐ no

Race: ☐ American Indian/Alaska Native ☐ Asian ☐ Black/African American ☐ Native Hawaiian/other Pacific Islander ☐ White

MRN/Case # | DOC # | Outbreak # | Submitter Lab #

Date Collected: | Time Collected: ☐am ☐pm | *Vaccination History: _____ _____ _____ _____

Previous Test Done? ☐ no ☐ yes Name of Test_____ Date _____ ☐ 1st ☐ 2nd ☐ 3rd State Lab Number: _____
Name of Test_____ Date _____ ☐ 1st ☐ 2nd ☐ 3rd State Lab Number: _____

Onset Date:_____ Exposure Date:_____ ☐ Clinical Illness/Symptoms:_____

⬇ SPECIMEN SOURCE CODE

Arbovirus Panels (Serum or CSF)

Mandatory: Onset Date, Collection Date, and Travel History

☐ Arbovirus Endemic Panel (WNV, EEE, SLE, LAC)

☐ Arbovirus Travel-Associated Panel (Chikungunya, Dengue)

Based on information provided PCR and/or immunological assays will be performed.

Required information, check all that apply:
DIAGNOSIS: ☐Aseptic Meningitis ☐Encephalitis ☐other _____

SYMPTOMS: ☐headache ☐fever ☐stiff neck ☐altered mental state ☐muscle weakness ☐rash ☐other _____

ILLNESS FATAL? ☐yes ☐no

TRAVEL HISTORY (dates and places)

_____

IMMUNIZATIONS: Yellow fever? ☐yes ☐no
Flavivirus? ☐yes ☐no

IMMUNOCOMPROMISED? ☐yes ☐no

☐ Aspergillus
☐ Chlamydia (group antigen IgG)
☐ Cryptococcal (antigen)
☐ Cytomegalovirus (CMV)
☐ Ehrlichia
☐ Epstein-Barr Virus (EBV)
☐ Hepatitis A Screen (IgM Ab only, acute infection)
Call lab (443-681-3889) prior to submitting
☐ Hepatitis B Screen (HBs antigen only)
Prenatal patient? ☐ yes ☐ no
☐ *Hepatitis B Panel: (HBsAg, HBsAb)
☐ *Hepatitis B post vaccine (HBsAb)
☐ Hepatitis C screen (HCV Ab only)

⬇ SPECIMEN SOURCE CODE

☐ Herpes Simplex Virus (HSV) Types 1&2
☐ Legionella
☐ Leptospira
☐ Lyme Disease
☐ *MMRV Immunity Screen: [Measles (Rubeola), Mumps, Rubella, Varicella (Chickenpox) IgG Ab only]
☐ Mononucleosis - Infectious
☐ *Mumps Immunity Screen
☐ Mycoplasma
☐ Rocky Mountain Spotted Fever (RMSF)
☐ *Rabies (RFFIT) (*List vaccination dates above)
☐ *Rubella Immunity Screen
☐ *Rubeola (Measles) Immunity Screen
☐ Schistosoma
☐ Strongyloides
☐ Syphilis - Previously treated? ☐ yes ☐ no
☐ Toxoplasma
☐ Tularemia
☐ Varicella Immunity Screen
☐ VDRL **(CSF only)**
☐ CDC/Other Test(s)

Add'l Specimen Codes _____ _____ _____

Prior arrangements have been made with the following DHMH Labs Administration employee: _____

_____

_____

_____

Please Note Vaccination History above*

⬇ SPECIMEN SOURCE CODE

⬇ LAVENDER TOP TUBE REQUIRED

☐ Hemoglobin Disorders

Blood transfusion? (last 4 months) ☐ yes ☐ no
Prenatal screen? ☐ yes ☐ no
Father of baby screen? ☐ yes ☐ no
Guardian's name if patient is a minor: _____
Name of mother of "at risk" baby:
_____

RESTRICTED TEST
Pre-Approved Submitters Only
Submit a separate specimen for HIV
Instructions go to:
http://dhmh.maryland.gov/laboratories/

☐ HIV
Country of Origin _____
Rapid Test: ☐ Reactive ☐ Negative
Date: _____

Specimen stored refrigerated (2°-8°c) after collection. ☐ yes ☐ no
Specimen transported on cold packs ☐ yes ☐ no

SPECIMEN SOURCE CODE:
PLACE CODE IN BOX NEXT TO TEST

B	**Blood** (5 ml)
CSF	**Cerebrospinal Fluid**
L	**Lavender Top Tube**
P	**Plasma**
S	**Serum** (1 ml per test)
UR	**Urine**